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Effect of Varying Rest Intervals Between Sets of Assistance Exercises on Creatine Kinase and Lactate Dehydrogenase Responses


ABSTRACT

Effect of varying rest intervals between sets of assistance exercises on creatine kinase and lactate dehydrogenase responses. J Strength Cond Res 25(5): 1339-1345, 2011-To examine the effects of different rest intervals between sets on serum creatine kinase (CK) and lactate dehydrogenase (LDH) activity, 10 men (age = 25.6 ± 2.2 years, height = 173.1 ± 7.1 cm, and body mass = 75.9 ± 10.0 kg) participated in a randomized within-subject design that involved 4 resistance exercise sessions. Each session consisted of 4 sets of 10 repetitions with 10 repetition maximum loads for the chest press, pullover, biceps curl, triceps extension, leg extension, and prone leg curl. The sessions differed only in the length of the rest interval between sets and exercises, specifically: 60, 90, 120, 180 seconds. Serum CK and LDH were significantly \( (p < 0.05) \) elevated 24-72 hours after each session, with no significant differences between rest intervals \( (p = 0.94 \text{ and } p = 0.99, \text{ respectively}) \). The mechanical stress imposed by the 4 resistance exercise sessions invoked similar damage to the muscle fibers independent of the rest interval between sets. These data indicate that the accumulated volume of work is the primary determinant of muscle damage in trained subjects who are accustomed to resistance exercise with short rest intervals.

INTRODUCTION

Resistance training can increase absolute strength, hypertrophy, muscular power, and localized muscular endurance, all characteristics that can contribute to improvements in physical function and quality of life. Several variables can be manipulated to address these characteristics, including muscle action, intensity (load), volume, exercise selection, exercise order, frequency of exercise sessions, velocity of muscle action, and rest intervals between sets (1,11,22,26).

The rest interval between sets has a critical role in resistance exercise prescription. Anecdotally, the rest interval between sets may not be monitored as closely as other variables (e.g., intensity and volume), despite significant effects on metabolic, hormonal, and cardiovascular responses (11,13,26). Shorter rest intervals between sets resulted in significant increases in epinephrine, norepinephrine, cortisol, and growth hormone (5,10,13). These hormones all play a role in the immunological response that occurs after heavy resistance exercise (18), as demonstrated by concomitant elevations in prostaglandin E2, tumor necrosis factor-\( \alpha \), interleukin 1b, interleukin 6, and interferon-\( \alpha \) (2,24). Therefore, shorter rest intervals between sets may invoke a significantly greater immune response vs. longer rest intervals between sets (14).
Serum levels of muscle enzymes serve markers of the status of skeletal muscle tissue and vary widely in both pathological and physiological conditions. Increases in the serum levels of these enzymes may represent an index of cellular necrosis and tissue damage after strenuous exercise and might be a consequence of both metabolic and mechanical stimuli. Indeed, metabolically exhausted muscle fibers exhibit an increase in membrane permeability consequent to an increase in free calcium ions, which promotes the activation of potassium channels and proteolytic enzymes (calpaines, caspases, etc.). Another mechanism could be the mechanical disruption and degeneration of the sarcomere from Z-disk fragmentation (4,16).

Serum creatine kinase (CK) and lactate dehydrogenase (LDH) have been studied extensively as markers for skeletal muscle microtrauma (6,8,17,19,24). Serum CK activity increases to a greater extent vs. the serum activity of other muscle proteins (e.g. LDH, myoglobin). As a result, serum CK activity is used widely as a marker of the status of muscle tissue (4,8,16,17). Few studies have been conducted to compare the differences in serum CK and LDH activity with different rest intervals between resistance exercise sets.

Mayhew et al. (14) compared serum CK activity after 2 bouts that consisted of 10 sets of 10 repetitions at 65% of 1 repetition maximum (1RM) in the leg press with either 1 or 3-minute rest intervals between sets. Significant elevations in serum CK activity were demonstrated at 24 hours postexercise for both rest conditions. However, serum CK activity was significantly greater for the 1-minute bout. Conversely, Ribeiro et al. (20) compared serum CK activity after performance of a total-body session that consisted of 3 sets of 10 repetitions with 10RM loads for the bench press, pullover, military press, biceps curl, triceps extension, leg press, leg extension, and leg curl with either 1- or 3-minute rest intervals between sets. However, in contrast to Mayhew et al. (14), Ribeiro et al. (20) demonstrated similar serum CK activity 24–48 hours postexercise for both rest conditions.

A possible explanation for this apparent difference is that Mayhew et al. (14) equalized the volume (load x sets x repetitions) completed between the 1- and 3-minute bouts, whereas Ribeiro et al. (20) did not, so that subjects completed a greater volume (load x sets x repetitions) for the 3-minute condition. More recently, Rodrigues et al. (21) reported similar CK and LDH activity after resistance exercise bouts with either 1- or 3-minute rest intervals between sets, and subjects completed 24% greater volume (load 3 sets 3 repetitions) for the 3-minute bout. Thus, it appears that postexercise CK and LDH activities might be related to both the volume (load x sets x repetitions) completed and the rest interval between sets.

Based on these inconsistencies, there is a need for more research to determine the effect of different rest intervals on serum CK and LDH activities. Specifically, there is a need to examine CK and LDH activities with a wider spectrum of rest intervals and over longer postexercise recovery periods. Therefore, the purpose of the current
study was to examine the effects of multiple rest intervals between sets and exercises (i.e., 60, 90, 120, and 180 seconds) on serum CK and LDH activities from 24 to 72 hours after performance of a totalbody resistance exercise bout. We hypothesized that shorter (60- and 90-second) rest intervals would lead to greater postexercise elevations in enzymes vs. longer (120- and 180- second) rest intervals.

METHODS

Experimental Approach to the Problem

Four resistance exercise bouts were performed using a randomized within subjects design. Each bout consisted of 6 resistance exercises, all of which could be classified as “assistance exercises” because of the relatively small muscle mass they activate; a similar exercise bout was used previously to examine the effect of training status on the CK response to resistance exercise (15). Before the intervention, 2 familiarization sessions (separated by 72 hours) were conducted to determine 10RM loads for all exercises and also collect anthropometric variables. Seven days after the last familiarization session, subjects performed the first of 4 resistance exercise bouts (each separated by 7 days) that consisted of 4 sets of each exercise with the 10RMload and 60-, 90-, 120-, or 180-second rest between sets and exercises. Serum CK and LDH activities were measured pre-exercise (T0), 24, 48, and 72 hours postexercise (T24, T48, and T72).

Subjects

Ten healthy men with resistance training experience volunteered to participate in this study. Before the commencement of the experiment, subjects had been participating in resistance training for a minimum of 12 months with a mean frequency of 3 sessions per week, and using approximately 1- to 2-minute rest intervals between sets and exercises. Subjects were healthy (no muscle, cardiovascular or joint problems) and were not using ergogenic substances or any other drugs. All procedures were approved by the local Ethics Committee. The purpose and procedures were explained to the subjects and informed consent was obtained according to the Declaration of Helsinki and in accordance with the norms of the local Ethics Committee.

Procedures

Before the intervention, 2 familiarization sessions were conducted to determine 10RMloads for the chest press, pullover, biceps curl, triceps extension, leg extension, and prone leg curl (Righetto, Brazil). A major consideration in choosing these exercises was the relative ease with which testers could ensure that subjects were performing them with a consistent range of motion. Given the repeated measures nature of the experiment, we felt it was important to ensure that all sessions were performed with consistent exercise technique, so that there were no
differences in work completed between each session. To increase the reliability of 10RM testing, the following strategies were employed: (a) the 10RM for each exercise was measured on 2 nonconsecutive days that were separated by 72 hours, (b) exercise testing proceeded in the same sequence as listed above, (c) exercise technique was monitored and corrected as needed, and (d) all subjects received verbal encouragement.

One week after the last familiarization session, subjects performed the first resistance exercise bout with either 60- (RI60), 90- (RI90), 120- (RI120), or 180- (RI180) second rest between sets and exercises. Warm-up before each bout consisted of pedaling 10 minutes on a cycle ergometer and dynamic stretching of the involved muscle groups. The repetition cadence for each exercise was controlled with a digital sound signal (Beat Test & Training, CEFISE, São Paulo, Brazil) that was adjusted so that each repetition was completed in approximately 2 seconds. A spotter gave minimal assistance if necessary so that 10 repetitions were completed on all 4 sets for each exercise. Therefore, the volume (load 3 sets 3 repetitions) completed was equalized between the resistance exercise bouts.

Subjects provided blood samples in a seated position from an antecubital vein into plain evacuated tubes in the morning after an overnight fast pre-exercise (T0), and 24 (T24), 48 (T48), and 72 (T72) hours postexercise. The serum was quickly frozen and stored at 270°C. From the serum sample, CK and LDH activity was measured. An enzymatic method was used for enzyme activity analysis with commercial kits (BioTecnica - Brazil) in Cobas Mira Plus analyzer (Roche, Basel, Switzerland). The same methodology was used in Machado et al (12) and that there was high reproducibility between measurements (intraclass coefficient r = 0.99).

**Statistical Analyses**

Multivariate analysis of variance (MANOVA) with repeated measures was used to test differences between time points and rest interval conditions. The alpha level was set at less than 0.05 for a difference to be considered significant. Significant main effects were further analyzed using pairwise comparisons with Tukey's post hocs. Effect sizes were computed using an Eta2. Statistical analysis was completed using SPSS_ 15.0 for Windows (LEAD Technologies, Haddonfield, NJ, USA).

**RESULTS**

The group studied exhibited little variability in demographical characteristics and high reliability for the 10RM exercise assessments (see Tables 1 and 2). The repeated measures MANOVA revealed no significant interaction between rest interval length and measurement time for either CK (p = 0.845, Eta2 = 0.46) or LDH (p = 0.996, Eta2 = 0.26) activity. However, both CK (p, 0.001, Eta2 = 2.82) and LDH (p, 0.001, Eta2 = 4.44) were significantly elevated above pre-exercise levels at 24, 48, and 72 hours (Figure 1). Individual responses for CK and LDH to each of the exercise bouts are displayed in Figures 2 and 3, respectively.
Figure 1. Creatine kinase (CK) and lactate dehydrogenase (LDH) concentrations measured before (T0), 24-hour (T24), 48-hour (T48), and 72-hour (T72) postresistance exercise bouts of with different rest interval lengths. Rest intervals were 60 seconds (RI60), 90 seconds (RI90), 120 seconds (RI120), and 180 seconds (RI180) between sets. *Significantly different from PRE value; p < 0.05.
Figure 2. Creatine kinase (CK) individuals’ response measured before (T0), 24-hour (T24), 48-hour (T48), and 72-hour (T72) postresistance exercise bouts. A) RI60; B) RI90; C) RI120; and D) RI180.

Figure 3. Lactate dehydrogenase (LDH) individuals’ response measured before (T0), 24-hour (T24), 48-hour (T48), and 72-hour (T72) postresistance exercise bouts. A) RI60; B) RI90; C) RI120; and D)–RI180.
DISCUSSION

Serum CK and LDH activities commonly serve as markers of the status of muscle fiber membranes after bouts of strenuous exercise. Serum CK activity has been shown to be elevated for 24 hours after exercise bouts, with a gradual return to basal levels in 72–96 hours (4,16). Serum LDH activity has been shown to be elevated 24 hours after exercise bouts and is maintained for 48–72 hours (3,7). The current study corroborates with previous investigations, in that CK and LDH activities were significantly elevated above pre-exercise levels at 24, 48, and 72 hours postexercise.

In the current study, the absolute values for LDH activity were modestly higher when compared with CK activity at all time points (time effect $\eta^2 = 4.44$ vs. $\eta^2 = 2.82$ for LDH and CK, respectively). However, as expected, the smaller CK protein ($\sim $86 kDa) displayed a greater proportional increase from the resting (T0) sample in all postexercise samples than did the larger LDH protein ($\sim $140 kDa). These data are corroborated by many other studies (3).

Previous studies that assessed CK and LDH after resistance exercise used 1 exercise, which has limited application for typical resistance exercise bouts that involve multiple exercises (4,8,17). Therefore, the design of the current study was more applicable to practical scenarios and demonstrated that there were no significant differences in CK or LDH activity between different rest interval conditions. This was a surprising finding given that in previous studies different rest intervals between sets and exercises elicited differences in physiological, biochemical, and hormonal responses (10,13,26).

Mayhew et al. (14) compared serum CK activity after 2 sessions that consisted of 10 sets of 10 repetitions at 65% 1RM in leg press with either 1- or 3-minute rest intervals between sets. Significant elevations in serum CK activity were demonstrated at 24 hours postexercise for both rest conditions. However, serum CK activity was significantly greater for the 1-minute bout. The differences between the findings of the current study vs. Mayhew et al. might be because of the intensity and number of exercises included in the exercise bout, both factors being greater in the current study.

But perhaps a more likely explanation for the lack of an effect of different rest intervals on the appearance of circulating enzymes in the current study is the training status of the subjects (25). Previous studies on the same topic used subjects who were experienced with resistance training, but the rest interval length with which they were accustomed to when training was not recorded (14,21). When timed rest periods are not enforced, both trained and untrained subjects have been found to rest approximately 3 minutes between sets (23). Thus, it is likely that the subjects in previous studies were not accustomed to training with shorter rest intervals (e.g., 1 minute).
In the current study, subjects did have a history of resistance training with shorter rest intervals between sets (e.g., 1–2 minutes) before their participation in the experiment. Consequently, they had likely already adapted to this type of training. Previous studies have consistently documented that resistance exercise with shorter rest intervals between sets (e.g., 1 minute) produces greater increases in circulating hormones vs. resistance exercise with longer rest intervals between sets (e.g., 3 minutes) (11,13,26). However, these studies are typically cross-sectional comparisons, using subjects with little or no training experience.

Buresh et al. (5) recently measured the hormonal response to resistance exercise between groups employing short (1 minute) vs. long (2.5 minutes) rest intervals longitudinally, over the course of a 10-week training cycle. Although the short rest interval group displayed a significantly higher elevation in postexercise testosterone and cortisol than was seen in the long rest interval group after 1 week of training, these differences had disappeared by 5 weeks of training. Thus, it appears that subjects can quickly adapt to training at a specific rest interval, after which postexercise increases in circulating hormones (and perhaps enzymes) will depend less on the rest interval employed and more on the total volume (load 3 sets 3 repetitions) of work completed. Indeed, such an adaptation would also explain the findings of Ribeiro et al. (20), who, in agreement with the current study, found no significant differences in CK or LDH activity after resistance exercise with either 1- or 3-minute rest intervals. Subjects in Ribeiro’s study, similar to those in the current study, were accustomed to training with 1- to 2-min rest intervals between sets before the experiment.

Our findings are further supported by the recent work of Hudson et al. (9). They examined oxidative stress markers in response to 2 resistance exercise protocols. In their study, subjects completed 2 bouts of back-squat exercise in a randomized order. One bout was a hypertrophy protocol (4 3 10 repetitions at 75% 1RM with 90-second rest intervals), the other bout was a strength protocol (11 3 3 repetitions at 90% 1RM with 5-minute rest intervals). The 2 bouts were equalized in terms of the total amount of work (Joules) completed. Similar to the present study, Hudson et al. (9) originally hypothesized that the hypertrophy bout, partly because of its shorter rest intervals, would elicit a higher oxidative stress response. However, like us, they were surprised to find that there were no differences in the presence of plasma biomarkers of oxidative damage (lipid hydroperoxides and protein carbonyls) between the bouts. The subjects in Hudson et al.’s study (9) were well trained before the experiment (average back squat 1RM = 1.9 3 body mass), although no specific mention was made of the rest intervals with which they were accustomed to training. Based on these findings, Hudson et al. (9) concluded that moderate (hypertrophy)- or high (strength)-intensity back squats yield similar oxidative damage responses. Although our study was limited to the measurement of serum enzymes as markers of muscle damage, we expected free radicals to contribute to the extent of muscle damage and that, in part, was why we hypothesized that shorter rest intervals would induce higher levels of CK and LDH in circulation. When we found similar CK and LDH levels, regardless of rest
interval, it was suggested that perhaps our choice of assistance exercises was an insufficient central or local metabolic stress to induce enough chemical damage from free radicals with the shorter rest intervals. Hudson et al.’s (9) findings, based on a back-squat exercise protocol, provide strong confirmation to our conclusion that rest interval length is not a determining factor in the extent of postexercise muscle damage.

In the present study, the mechanical stress imposed by the resistance exercise sessions invoked similar damage to the muscle fibers independent of the rest interval between sets. These data indicate that the accumulated volume (load3sets 3 repetitions) of work is the primary determinant of muscle damage in trained subjects who are accustomed to resistance exercise with short rest intervals.

**PRACTICAL APPLICATIONS**

The data from the current study would be useful when prescribing resistance exercise programs to allow for sufficient recovery between training sessions. If recreationally trained men are performing multiple sets of full repetition maximums, as may be done during high-volume hypertrophy mesocycles of a periodized plan, greater than 3 days (i.e., 72 hours) between workouts for the same muscle groups might be necessary to allow for sufficient muscle repair, recovery, and ultimately adaptation. However, if the men are accustomed to training with short rest intervals, the length of rest interval between sets does not appear to have any effect on the extent of muscle damage. Thus, during high-volume, low-intensity resistance training, exercising with short rest intervals does not appear to present any additional challenge to recovery.

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